IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

#36
Linda 7/31/03

Appellants: Eugen Koren and Mirna Koscec

Serial No.: 08/970,045 Group Art Unit: 1645

Filed: November 13, 1997 Examiner: P. Duffy

For: *ANTIBODIES TO LIPOPROTEINS AND APOLIPOPROTEINS AND METHODS OF USE THEREOF*

Assistant Commissioner for Patents
Washington, D.C. 20231

APPEAL BRIEF

Sir:

This is an appeal from the rejection of claims in the Office Action mailed October 2, 2002, maintained in the Advisory Action mailed February 6, 2003, and further to the Decision on Petition dated May 13, 2003. The Commissioner is authorized to charge the fee for this Appeal Brief of $160.00 to Deposit Account No. 50-1868.

It is believed that no additional fee is required with this submission. However, should a fee be required, the Commissioner is hereby authorized to charge the fee to Deposit Account No. 50-1868.

(1) REAL PARTY IN INTEREST

The real party in interest of this application is the assignee, Oklahoma Medical Research Foundation, Oklahoma City, OK., and the licensee, Sigma Chemical Co, now doing business as Sigman-Aldrich Inc.

(2) RELATED APPEALS AND INTERFERENCES

There are no related appeals or interferences known to appellant, the undersigned, or appellant's assignee which directly affects, which would be directly affected by, or which would have a bearing on the Board's decision in this appeal.

(3) STATUS OF CLAIMS ON APPEAL

Claims 1-13, 39-47 are pending. Claims 1-11, 39, 41-43, 46 and 47 are allowed. Claims 44 and 45 should be allowed in view of the accompanying amendment. Claims 12, 13, and 40 are on appeal. The text of each claim, as pending, is set forth in an Appendix to this Appeal Brief.

(4) STATUS OF AMENDMENTS

The claims were last amended in the appellant's response received on April 22, 2002. An amendment accompanies this Appeal Brief.

(5) SUMMARY OF THE INVENTION

Compositions and methods using antibodies which are immunoreactive with specific apolipoproteins to determine the concentrations of lipoproteins such as HDL and LDL, and/or apolipoproteins in human blood, serum or plasma sample, are described (this paragraph is found in the application at page 14, line 17 to page 15, line 21). Monoclonal antibodies (MAbs) are described that specifically bind to epitopes present in apolipoproteins and lipoproteins, enabling rapid and reliable determinations of levels of specific blood lipoprotein and/or apolipoprotein levels, including Apo B-100, Apo A-I, Apo A-II, Apo C-III, and Apo E, and thereby determination of relative ratios of HDL and LDL and LpaI and LpaII. In a preferred embodiment, the compositions are strips of a solid phase material coated with one or more of the

antibodies and are referred to herein as "dipsticks". The dipsticks specifically bind a lipoprotein or apolipoprotein when dipped into a protein sample. The amount of lipid associated with a bound lipoprotein or the amount of apolipoprotein bound on the dipstick is quantitated using an appropriate method, for example, by staining with a lipid stain or reaction with a second labelled antibody. The intensity of the stain on the dipstick is proportional to the concentration of the lipoprotein lipid or apolipoprotein circulating in the blood and can be quantitated by comparison with standards containing known amounts of lipid. The dipsticks can be provided alone or in kits which enable the lay person to carry out the assay without the need of a physician or technical laboratory..

Claims 12 and 13 define a method of determining the relative concentration of at least two different apolipoproteins in a biological sample comprising:

mixing in solution a first and second monoclonal antibody molecules, each immunoreactive with a specific different apolipoprotein into the sample, wherein at least one of the first and second monoclonal antibodies bind to a stable, conformation independent epitope of a lipoprotein that is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with the specific lipoprotein in a conformation and lipid content independent manner;

allowing the monoclonal antibody molecules to bind to the apolipoproteins in the sample,

immersing into the mixture third immobilized monoclonal antibody molecules immunoreactive with a second, distinct epitope of one of the first or second apolipoproteins,

allowing the third immobilized monoclonal antibody molecules to bind to one of the apolipoproteins bound by either the first or second monoclonal antibodies,

determining the amount of apolipoprotein bound by the first and second monoclonal antibodies and the amount of protein bound by the third immobilized monoclonal antibodies, and

subtracting from the total apolipoprotein bound by the first and second monoclonal antibodies the amount of protein bound by the third immobilized monoclonal antibodies, to yield the amounts of the first and second apolipoproteins.

Claim 40 defines a method for determining the relative ratio of VLDL to HDL in a biological sample comprising

(a) determining the amount of VLDL in the sample by

determining the amount of Apo C-III present in the VLDL in the sample by

providing Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

providing monoclonal antibody specifically immunoreactive with Apo C-III,

contacting the anti-ApoC-III antibody reactive with Apo C-III with the biological sample to form complexes between the anti-ApoC-III antibody and the Apo C-III containing lipoprotein particles,

contacting the Pan B antibody with the biological sample containing the anti-ApoC-III antibody bound to the Apo C-III containing lipoprotein particles,

separating the complexed Pan B-anti-ApoC-III antibody-lipoprotein particles from the biological sample, and

determining the amount of complexed Pan B-anti-ApoC-III antibody-lipoprotein particles, which is the amount of Apo C-III present in VLDL in the anti-Apo C-III anti-Apo B complexed material in the sample; and

(b) determining the amount of HDL in the sample by

determining the amount of Apo C-III present in the HDL in the sample by

providing Apo A-I monoclonal antibody specifically immunoreactive with Apo A-I,

providing monoclonal antibody specifically immunoreactive with Apo C-III,

contacting the antibody reactive with Apo C-III with the biological sample to form complexes between the anti-Apo C-III antibody and the Apo C-III containing lipoprotein particles,

contacting the anti-Apo A-I antibody with the biological sample to form complexes with the anti-Apo C-III antibody-Apo C-III containing lipoprotein particles,

separating the complexed anti-Apo C-III antibody-Apo C-III containing lipoprotein particles from the biological sample,

determining the amount of Apo C-III present in HDL in the anti-Apo C-III-anti-Apo A-I complexed material in the sample, and

determining the ratio of Apo C-III present in VLDL in the sample to Apo C-III present in HDL in the sample, which is the ratio of VLDL to HDL,

wherein the VLDL and HDL are measured in the same sample using immobilized anti-Apo A-I and anti-Apo B or anti-Apo C-III antibodies or measured by immunoprecipitation with the anti-Apo A-I and anti-ApoB antibodies or anti-Apo C-III antibodies in separate samples,

wherein at least one of the monoclonal antibodies bind to a stable, conformation independent epitope that is uninfluenced by the lipid content of the lipoprotein, apolipoprotein or lipid associated with a specific lipoprotein selected from the group consisting of Apo AI, Apo B, and Apo CIII.

(6) ISSUES ON APPEAL

The issues presented on appeal are:

(1) whether claims 12, 13, and 40 are indefinite under 35 U.S.C> 112, second paragraph, as indefinite.

(7) GROUPING OF CLAIMS

Claims 12 and 13 stand or fall together. Claim 40 stands alone. The issues discussed below are specific to the claims, not to the group.

(8) ARGUMENTS

(a) The Claimed Invention

The claimed invention are assays which utilize antibodies to lipoproteins having a unique reactivity to an epitope that is independent of the lipid concentration (which may vary drastically in the case of lipoproteins) and conformation. These assays utilize antibodies to other epitopes or antigens which can be used for comparison, in order to determine relative concentrations. For example, the allowed claim 1 defines an assay as follows:

A method for determining the relative ratio of at least two different lipoproteins or apolipoproteins in a biological sample comprising:

immersing into the sample a solid phase material having separately immobilized thereon at least first and second antibody molecules, wherein the antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies and antigen-binding antibody fragments thereof, wherein the antibody molecules are immunoreactive with at least two different lipoproteins, ***wherein the first and second antibodies bind to different stable, conformation independent epitopes that are uninfluenced by the lipid content of the***

lipoprotein, protein component of the lipoprotein or lipid associated with the specific lipoprotein, wherein the lipoproteins are selected from the group consisting of LDL, HDL and VLDL;

allowing the antibody molecules time to bind to the LDL, HDL, VLDL or apolipoproteins in the sample;

removing the solid phase material containing the immobilized antibody molecules;

determining the amount of lipoprotein or apolipoproteins bound by the immobilizedantibody molecules, and

comparing the amount bound which is specific for LDL, HDL, VLDL or each apolipoprotein in order to calculate the relative amounts of LDL, HDL, VLDL or apolipoproteins.

The advantage of these types of assays is that they allow one to use a single dipstick or turbidometric assay to measure multiple criteria at the same time. They also allow measurement of samples directly, without pre-treatment to remove lipid and cross-reactive materials.

There are two independent claims in issue: claims 12 and 40, where the examiner has alleged that the claims are indefinite.

(b) Rejections Under 35 U.S.C. § 112, second paragraph

i. Legal Standard for Definiteness

Definiteness problems arise most often when words of degree, relational terms, and ranges are used in claims language. Frequently at issue are words such as ``substantially," 81. ``relatively," and ``closely." Such concepts do not render a claim fatally indefinite if the specification provides a standard for measuring substantiality, relativity or closeness such that

one skilled in the art can determine whether a particular product or process falls within the language of the claim. The courts clearly are influenced by their perception of whether the patentee has been ``as precise as the subject matter permits" or rather has tried ``to corral the art by the use of comprehensive indefinite terms."

In Rengo Co. Ltd. v. Molins Mach. Co. (1981), the Third Circuit noted a ``subtle relationship between the policies underlying the description and definiteness requirements." ``[T]he two standards, while complementary, approach a similar problem from different directions. Adequate description of the invention guards against the inventor's overreaching by insisting that he recount his invention in such detail that his future claims can be determined to be encompassed within his original creation. The definiteness requirement shapes the future conduct of persons other than the inventor, by insisting that they receive notice of the scope of the patented device." McClain v. Ortmayer, 141 U.S. 419, 424 (1891), ``The object ... is not only to secure to [the patentee] all to which he is entitled, but to apprise the public of what is still open to them."; Solomon v. Kimberly-Clark Corp., 216 F.3d 1372, 55 USPQ2d 1279 (Fed. Cir. 2000) (quoting Treatise); Mycogen Plant Science, Inc. v. Monsanto Co., 61 F. Supp. 2d 199, 255 (D. Del. 1999) (citing Treatise); Austin Powder Co. v. Atlas Powder Co., 568 F. Supp. 1294, 219 USPQ 707 (D. Del. 1983) (citing Treatise). United Carbon Co. v. Binney Co., 317 U.S. 228, 236 (1942).

The claims are to be interpreted first based on the actual language of the claims, using the standard meaning of the words, then within the context of the specification.

ii. Claims 12 and 13.

The examiner has alleged that claim 12, and claim 13 dependent thereon, is indefinite on the basis that the preamble and the method steps do not match.

Claim 12 recites:

A method of determining the relative concentration of at least two different apolipoproteins in a biological sample comprising:

(1) mixing in solution a first and second monoclonal antibody molecules, each immunoreactive with a specific different apolipoprotein into the sample, wherein at least one of the first and second monoclonal antibodies bind to a stable, conformation independent epitope of a lipoprotein that is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with the specific lipoprotein in a conformation and lipid content independent manner;

(2) allowing the monoclonal antibody molecules to bind to the apolipoproteins in the sample,

(3) immersing into the mixture third immobilized monoclonal antibody molecules immunoreactive with a second, distinct epitope of one of the first or second apolipoproteins,

(4) allowing the third immobilized monoclonal antibody molecules to bind to one of the apolipoproteins bound by either the first or second monoclonal antibodies,

(5) determining the amount of apolipoprotein bound by the first and second monoclonal antibodies and the amount of protein bound by the third immobilized monoclonal antibodies, and

(6) subtracting from the total apolipoprotein bound by the first and second monoclonal antibodies the amount of protein bound by the third immobilized monoclonal antibodies, to yield the amounts of the first and second apolipoproteins.

The claim is quite definite. Steps 1 and 2 yield apolipoprotein 1 bound to one antibody (1a) and apolipoprotein 2 bound to a second antibody (2a).

Steps 3 and 4 yield either apoliprotein 1 or apolipoprotein 2 bound to a third antibody (either 1b or 2b).

Step 5 yields the concentration of apolipoprotein 1 plus apolipoprotein 2. Step 6 subtracts the amount of either apolipoprotein 1 or apolipoprotein 2 from the total, thereby yielding values for the total apolipoprotein 1 and total apolipoprotein 2.

The preamble provides for determining the relative concentrations of the two apolipoproteins. There is nothing inconsistent about the method steps and the preamble. The values are relative since they are determined by reference to each other.

Figuratively:

Apo1<->AbApo1a Apo2<->AbApo2a

Apo1<->AbApo1a OR ApoA2<->Apo2a
↕ ↕
AbApo1b AbApoA2b

Total of Apo1 plus Apo2
Subtract
Total of EITHER Apo1a/Apo1b OR Apo2a/Apo2b

=

in the first alternative, the amount of Apo1 will be that bound by Ab to Apo1a and Apo1b and the difference will be the amount of Apo2

in the second alternative, the amount of Apo2 will be that bound by Ab to Apo2a and Apo2b, and the difference will be the amount of Apo1.

In summary, the claim is definite as required under 35 U.S.C. 112. One of ordinary skill in the art would have no trouble understanding what is claimed.

With respect to where in the specification there is support, the specifaction stated "determining the difference between" in example 10, page 66, line 31, to page 68, line 11. One skilled in the art would know that "substracting" and "determining the difference between" are equivalent and would be understood by those skilled in the art as meaning the same thing.

iii. Claim 40

The examiner has rejected claim 40 on the basis that "separating the complexed anti-ApoC-III antibody ApoC-III containing lipoprotein particles from the biological sample" is unclear.

Claim 40 defines a method for determining the relative ratio of VLDL to HDL in a biological sample comprising

(a) determining the amount of VLDL in the sample by

determining the amount of Apo C-III present in the VLDL in the sample by

providing Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

providing monoclonal antibody specifically immunoreactive with Apo C-III,

contacting the anti-ApoC-III antibody reactive with Apo C-III with the biological sample to form complexes between the anti-ApoC-III antibody and the Apo C-III containing lipoprotein particles,

contacting the Pan B antibody with the biological sample containing the anti-ApoC-III antibody bound to the Apo C-III containing lipoprotein particles,

separating the complexed Pan B-anti-ApoC-III antibody-lipoprotein particles from the biological sample, and

determining the amount of complexed Pan B-anti-ApoC-III antibody-lipoprotein particles, which is the amount of Apo C-III present in VLDL in the anti-Apo C-III anti-Apo B complexed material in the sample;

and

(b) determining the amount of HDL in the sample by

determining the amount of Apo C-III present in the HDL in the sample by

providing Apo A-I monoclonal antibody specifically immunoreactive with Apo A-I,

providing monoclonal antibody specifically immunoreactive with Apo C-III,

contacting the antibody reactive with Apo C-III with the biological sample to form complexes between the anti-Apo C-III antibody and the Apo C-III containing lipoprotein particles,

contacting the anti-Apo A-I antibody with the biological sample to form complexes with the anti-Apo C-III antibody-Apo C-III containing lipoprotein particles,

separating the complexed anti-Apo C-III antibody-Apo C-III containing lipoprotein particles from the biological sample,

determining the amount of Apo C-III present in HDL in the anti-Apo C-III-anti-Apo A-I complexed material in the sample, and

determining the ratio of Apo C-III present in VLDL in the sample to Apo C-III present in HDL in the sample, which is the ratio of VLDL to HDL,

wherein the VLDL and HDL are measured in the same sample using immobilized anti-Apo A-I and anti-Apo B or anti-Apo C-III antibodies or measured by immunoprecipitation with the anti-Apo A-I and anti-ApoB antibodies or anti-Apo C-III antibodies in separate samples,

wherein at least one of the monoclonal antibodies bind to a stable, conformation independent epitope that is uninfluenced by the lipid content of the lipoprotein, apolipoprotein or lipid associated with a specific lipoprotein selected from the group consisting of Apo AI, Apo B, and Apo CIII.

Claim 40 is clear to one of ordinary skill in the art.

Step a yields a total of ApoCIII bound in VLDL, which is identified by being complexed with antibody to Apo B (i.e., the total VLDL is that lipoprotein bound by antibody to both ApoCIII and antibody to ApoB).

Step b yields a total of ApoCIII bound in HDL, which is identified by being complexed with antibody to ApoAI (i.e., the total HDL is that lipoprotein bound by antibody to both ApoCIII and antibody to ApoAI).

The relative ratio of VLDL to HDL is the ratio of ApoCIII determined in step a to the ApoCIII determined in step b.

To the extent the examiner's rejection is that appellants did not list every single step which might be encompassed within the claimed method, it is respectfully noted that claims are only to define that which is appellant's invention; not that which is known and routine. Although in some embodiments one might chose to separate the two complexes for the determinations in step a and step b, it is also possible to achieve determination using a solution method, where the different complexes are measured using automated assay equipment, and the

antibodies have different labels. The method by which the assay can be achieved is routine; the point of novelty and non-obviousness is the selection and use of the claimed antibodies.

In summary, claim 40 is also definite to one of ordinary skill in the art, as required under 35 U.S.C. 112.

(9) SUMMARY AND CONCLUSION

The language in claims 12, 13 and 40 is clear and definite to one of ordinary skill in the art, using the ordinary meaning of the words, particularly in view of the specification, Accordingly, all claims should now be allowable.

For the foregoing reasons, Appellant submits that the claims 1-25 are patentable.

Respectfully submitted,

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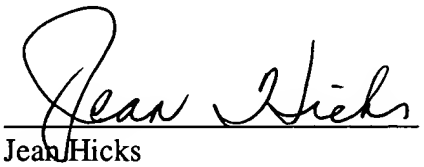

Jean Hicks

Date: July 11, 2003

APPENDIX: CLAIMS AS PENDING UPON ENTRY OF THE ACCOMPANYING AMENDMENT

1. (four times amended) A method for determining the relative ratio of at least two different lipoproteins or apolipoproteins in a biological sample comprising:

immersing into the sample a solid phase material having separately immobilized thereon at least first and second antibody molecules, wherein the antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies and antigen-binding antibody fragments thereof, wherein the antibody molecules are immunoreactive with at least two different lipoproteins, wherein the first and second antibodies bind to different stable, conformation independent epitopes that are uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with the specific lipoprotein, wherein the lipoproteins are selected from the group consisting of LDL, HDL and VLDL;

allowing the antibody molecules time to bind to the LDL, HDL, VLDL or apolipoproteins in the sample;

removing the solid phase material containing the immobilized antibody molecules;

determining the amount of lipoprotein or apolipoproteins bound by the immobilizedantibody molecules, and

comparing the amount bound which is specific for LDL, HDL, VLDL or each apolipoprotein in order to calculate the relative amounts of LDL, HDL, VLDL or apolipoproteins.

2. (amended) The method of claim 1 wherein the antibody molecules immobilized on the solid phase material are immunoreactive with lipoproteins selected from the group consisting of HDL and LDL.

3. (twice amended) The method of claim 2 wherein the antibodies to the HDL or LDL are selected from the group consisting of recombinant antibodies and antibody fragments.

4. (twice amended) The method of claim 3, wherein the first or second monoclonal antibodies are the anti-LDL monoclonal antibody produced by the hybridoma cell line HB_3cB_3 ATCC designation number HB 11612.

5. (twice amended) The method of claim 3, wherein the first or second monoclonal antibodies are recombinant anti-LDL $RcB_3M_1D_4$ ATCC designation number 69602.

6. (three times amended) The method of claim 1 further comprising determining the amount of lipoprotein lipid or lipid associating with apolipoprotein by staining of the material bound to the immobilized antibody using a lipid stain.

7. The method of claim 6 wherein the lipid stain is selected from the group consisting of Sudan Red 7B, Oil Red O, and Sudan Black B.

8. The method of claim 6 wherein the lipoprotein lipid is stained prior to immersing the immobilized antibodies.

9. (three times amended) The method of claim 6 further comprising measuring the amount of apolipoprotein or protein associated with the lipid in the sample, further comprising the step of providing antibodies immunoreactive with at least one apolipoprotein, wherein the antibodies are coupled to a protein stain, and staining the apolipoprotein or protein associated

with the lipid in the sample by reacting the protein stain coupled antibodies with the apolipoprotein or protein associated with the lipid in the sample.

10. The method of claim 1, wherein the apolipoprotein is selected from the group consisting of Apo A-I, Apo A-II, Apo B, Apo C-III, and Apo E.

11. The method of claim 1, wherein the biological sample is selected from the group consisting of blood, plasma, and serum.

12. (four times amended) A method of determining the relative concentration of at least two different apolipoproteins in a biological sample comprising:

mixing in solution a first and second monoclonal antibody molecules, each immunoreactive with a specific different apolipoprotein into the sample, wherein at least one of the first and second monoclonal antibodies bind to a stable, conformation independent epitope of a lipoprotein that is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with the specific lipoprotein in a conformation and lipid content independent manner;

allowing the monoclonal antibody molecules to bind to the apolipoproteins in the sample,

immersing into the mixture third immobilized monoclonal antibody molecules immunoreactive with a second, distinct epitope of one of the first or second apolipoproteins,

allowing the third immobilized monoclonal antibody molecules to bind to one of the apolipoproteins bound by either the first or second monoclonal antibodies,

determining the amount of apolipoprotein bound by the first and second monoclonal antibodies and the amount of protein bound by the third immobilized monoclonal antibodies, and

subtracting from the total apolipoprotein bound by the first and second monoclonal antibodies the amount of protein bound by the third immobilized monoclonal antibodies, to yield the amounts of the first and second apolipoproteins.

13. (amended) The method of claim 12 wherein the apolipoprotein bound by one of the monoclonal antibodies in solution is apolipoprotein Apo B-100.

39. (three times amended) A method for determining the relative ratio of LDL to HDL in a biological sample comprising

(a) determining the amount of LDL in the sample by

adding to the sample monoclonal antibody molecules immunoreactive with low density lipoprotein and not cross-reactive with high density lipoprotein and determining the amount of low density lipoprotein;

(b) determining the amount of HDL in the sample by

adding to the sample monoclonal antibody molecules immunoreactive with high density lipoprotein and not cross-reactive with low density lipoprotein and determining the amount of high density lipoprotein; and

(c) determining the ratio of the amount of low density lipoprotein with the amount of high density lipoprotein, wherein at least one of the monoclonal antibodies to LDL and HDL bind a stable, conformation independent epitope that is uninfluenced by the lipid content of the lipoprotein, the protein component of the lipoprotein or lipid associated with the specific lipoprotein.

40. (two times amended) A method for determining the relative ratio of VLDL to HDL in a biological sample comprising

(a) determining the amount of VLDL in the sample by

determining the amount of Apo C-III present in the VLDL in the sample by

providing Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

providing monoclonal antibody specifically immunoreactive with Apo C-III,

contacting the anti-ApoC-III antibody reactive with Apo C-III with the biological sample to form complexes between the anti-ApoC-III antibody and the Apo C-III containing lipoprotein particles,

contacting the Pan B antibody with the biological sample containing the anti-ApoC-III antibody bound to the Apo C-III containing lipoprotein particles,

separating the complexed Pan B-anti-ApoC-III antibody-lipoprotein particles from the biological sample, and

determining the amount of complexed Pan B-anti-ApoC-III antibody-lipoprotein particles, which is the amount of Apo C-III present in VLDL in the anti-Apo C-III anti-Apo B complexed material in the sample;

and

(b) determining the amount of HDL in the sample by

determining the amount of Apo C-III present in the HDL in the sample by

providing Apo A-I monoclonal antibody specifically immunoreactive with Apo A-I,

providing monoclonal antibody specifically immunoreactive with Apo C-III,

contacting the antibody reactive with Apo C-III with the biological sample to form complexes between the anti-Apo C-III antibody and the Apo C-III containing lipoprotein particles,

contacting the anti-Apo A-I antibody with the biological sample to form complexes with the anti-Apo C-III antibody-Apo C-III containing lipoprotein particles,

separating the complexed anti-Apo C-III antibody-Apo C-III containing lipoprotein particles from the biological sample,

determining the amount of Apo C-III present in HDL in the anti-Apo C-III-anti-Apo A-I complexed material in the sample, and

determining the ratio of Apo C-III present in VLDL in the sample to Apo C-III present in HDL in the sample, which is the ratio of VLDL to HDL,

wherein the VLDL and HDL are measured in the same sample using immobilized anti-Apo A-I and anti-Apo B or anti-Apo C-III antibodies or measured by immunoprecipitation with the anti-Apo A-I and anti-ApoB antibodies or anti-Apo C-III antibodies in separate samples,

wherein at least one of the monoclonal antibodies bind to a stable, conformation independent epitope that is uninfluenced by the lipid content of the lipoprotein, apolipoprotein or lipid associated with a specific lipoprotein selected from the group consisting of Apo AI, Apo B, and Apo CIII.

41. (three times amended) A method for determining the relative ratio of VLDL to HDL comprising

(a) determining the amount of VLDL in the sample by

determining the amount of Apo E present in the VLDL in the sample by

providing Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

providing monoclonal antibody which specifically binds to Apo E associated with VLDL,

contacting the antibodies reactive with Apo E associated with VLDL with the biological sample to form complexes between the anti-ApoE antibodies and Apo E containing particles,

contacting Pan B antibody with the biological sample containing the complexes between the anti-ApoE antibodies and ApoE containing particles to form complexes of anti-ApoB-anti-ApoE-ApoE containing particles, and

determining the amount of Apo E in the complexes of anti-ApoB-anti-ApoE-ApoE containing particles, which is the Apo E present in VLDL in the sample;

(b) removing the complexes of anti-ApoB-anti-ApoE-ApoE containing particles, either by binding of the anti-Apo E antibodies to an immobilized surface or centrifugation of sample to remove the complexes of anti-ApoB-anti-ApoE-ApoE containing particles;

and

(c) determining the amount of HDL in the sample by

determining the amount of Apo E present in the HDL in the sample by

providing Apo A-I monoclonal antibody immunoreactive specifically with Apo A-I,

contacting antibodies reactive with Apo E in HDL particles with the biological sample to form complexes between the anti-ApoE antibodies and Apo E containing particles,

contacting the Apo A-I monoclonal antibody with the biological sample to form complexes of the anti-ApoE antibodies-ApoE containing particles-anti-ApoA-I,

determining the amount of Apo E present in HDL in the complexes of the anti-ApoE antibodies-ApoE containing particles-anti-Apo A-I in the sample, and

determining the ratio of Apo E present in VLDL in the sample and Apo E present in HDL in the sample which is the ratio of VLDL to HDL,

wherein at least one of the monoclonal antibodies bind to a stable, conformation independent epitope that is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with a specific lipoprotein selected from the group consisting of Apo B, Apo AI, and Apo E.

42. (three times amended) A kit for determining the relative ratio of VLDL to HDL comprising

Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

monoclonal or recombinant antibody specifically immunoreactive with Apo C-III, and

monoclonal or recombinant Apo A-I antibody specifically immunoreactive with Apo A-I,

wherein at least one of the monoclonal or recombinant antibodies specifically bind to a stable, conformation independent epitope of a lipoprotein including Apo C-III or Apo A-I that is uninfluenced by the lipid content of the lipoprotein, protein component thereof or lipid associated with a specific lipoprotein selected from the group consisting of Apo AI, and Apo CIII.

43. (twice amended) The kit of claim 42 wherein the anti-Apo C-III or anti-A-1 monoclonal or recombinant antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies, and antigen binding antibody fragments thereof

that specifically bind to a stable, conformation independent epitope which is uninfluenced by the lipid content of the lipoprotein, protein component thereof, or lipid associated with a specific lipoprotein.

44. (presently three times amended) A kit for determining the relative ratio of VLDL to HDL comprising

Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

monoclonal antibody which binds to Apo E associated with VLDL ,

monoclonal Apo A-I antibody specifically immunoreactive with Apo A-I, and

monoclonal antibody which binds to Apo E in HDL,

wherein at least one of the antibodies binds to a stable, conformation independent epitope of a lipoprotein containing Apo E or Apo A-I that is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with a specific lipoprotein.

46. (twice amended) A kit for determining the relative ratio of LPA-I and LPA-II lipoprotein particles comprising

monoclonal or recombinant Apo-A-I antibody specifically immunoreactive with Apo A-I lipoproteins in human plasma; and

monoclonal or recombinant Apo A-II antibody specifically immunoreactive with Apo A-II,

wherein the anti-Apo A-I or anti-Apo A-II monoclonal or recombinant antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies, and antigen-binding antibody fragments thereof that specifically bind to a stable,

conformation independent epitope of a lipoprotein containing Apo A-I or Apo A-II which is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein, or lipid associated with a specific lipoprotein.

47. (twice amended) The kit of claim 46 wherein the anti-Apo A-I and anti-Apo A-II monoclonal or recombinant antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies, and monoclonal antibody fragments that specifically bind to a stable, conformation independent epitope which is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein, or lipid associated with a specific lipoprotein.

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